were used to calculate outcomes. The median age was 6.5 years, median cell dose infused was 4.1 × 10^7/kg and the median follow time was 22 months (3-96). The cord blood was HLA identical (6/6) in 12% of the cases, 5/6 in 46%, 4/6 in 39% and 3/6 in 3%. Overall 2 year-LFS was 36 ± 3%. In a multivariate analysis, only CR1 or CR2 were associated with better LFS (HR = 1.8; $P < 0.0001$). Therefore a separate analysis was performed according to the disease status. Disease and patient characteristics and outcomes are listed in the table. All children received myeloablative conditioning regimen and the majority (67%) received CsA+corticoids as GVHD prophylaxis.

In conclusion, these results confirm that UCBT should be proposed as alternative source of allogeneic transplantation for children with high risk ALL, in earlier status of the disease, for those children lacking an HLA identical donor.

23

IMPLEMENTING THE STEM CELL THERAPEUTIC AND RESEARCH ACT OF 2005

Baitty, Robert L., Tims, Shelley E., Wabeke, Anita, Ashton, Robyn, Gale, Randy, Burdick, James F. Department of Health and Human Services, Health Resources and Services Administration, Division of Transplantation, Rockville, MD.

The Stem Cell Therapeutic and Research Act of 2005 was signed by President Bush on December 20, 2005. Section 2 of the Act authorizes Federal support for collection of 150,000 new, high quality cord blood units for a National Cord Blood Inventory, authorizes a related cord blood demonstration program, and requires that some cord blood units not suitable for clinical transplantation be made available for research. Section 3, among other things, authorizes the C.W. Bill Young Cell Transplantation Program to facilitate unrelated blood stem cell transplants (as the successor to the National Bone Marrow Donor Registry), calls for establishment of an Advisory Council to advise the Secretary of the Department of Health and Human Services (HHS) regarding the Program and related matters, and requires HHS to recognize one or more accrediting organizations to accredit cord blood banks collecting units for the National Cord Blood Inventory. Administrative responsibility for most provisions of the Act is assigned to the Health Resources and Services Administration (HRSA) within the Department of Health and Human Services. This presentation describes HRSA's approach to implementing the new programs authorized by the Act, concentrating on the National Cord Blood Inventory and the Cord Blood Coordinating Center.

24

CULTURE OF CD34$^+$ UMBILICAL CORD BLOOD PROGENITORS WITH NOTCH LIGAND RESULTS IN ENHANCED AND MORE RAPID HUMAN ENGRAFTMENT IN A PRECLINICAL NOD/SCID MOUSE MODEL

Delaney, Colleen. Clinical Research Division, Fred Hutchinson Cancer Research Center, Seattle, WA, and Department of Pediatrics, University of Washington, Seattle, WA.

Our laboratory has developed novel methods for enhancing the magnitude and kinetics of hematopoietic reconstitution by human cord blood progenitor cells using the Notch ligand, Delta1. We have previously shown that culture of CD34$^+$CD38$^-$ UCB progenitors with Delta1 results in enhanced generation of NOD/SCID repopulating cells. Furthermore, we have examined effect of Notch ligand density on induction of endogenous Notch signaling and subsequent cell-fate of hematopoietic precursors, demonstrating critical quantitative aspects of Notch signaling in affecting cell fate outcome.

Based on these findings, we have developed a clinically feasible cGMP method for Notch ligand-based expansion of cord blood precursors. Specifically, we investigated the use of CD34$^+$ versus CD34$^+$CD38$^-$ cells as a starting population, optimal cytokines and medium, selection of culture vessel and culture period for effects on generation of NOD/SCID repopulating cells. UCB progenitors were cultured in the presence of a Notch ligand form consisting of the extracellular domain of Delta1 fused to the Fc domain of human IgG1 (Delta 1$^{ext\text{-}IgG}$) or control human IgG.

Initial studies demonstrated optimal cytokines consisted of SCF, FL, TPO, IL6 and IL3, together with fibronectin fragments in serum free medium. There was no significant difference seen in the CD34 fold expansion with CD34$^+$ versus CD34$^+$CD38$^-$ starting cells, however, upon transplantation into NOD/SCID mice, there was a significant increase in the level of human engraftment seen with the CD34$^+$ starting cell population (6.93% vs 2%; $P = 0.01$). Further results from 5 independent experiments in which cord blood CD34$^+$ progenitor cells were cultured for 17 days with immobilized Delta1^{extIgG} or control resulted in a mean fold expansion of CD34$^+$ cells of 230 (± 53) for the Delta cultured cells versus 65 (±31) for the control cultured cells ($P = 0.03$). Delta cultured cells demonstrated significantly enhanced levels of human engraftment as measured by percent CD45 in the marrow of the animals at both 3 weeks (Delta1 15.5%, control 2.6%, uncultured 0.2%; $P < 0.0001$) and at 9 weeks (Delta1 29.4%, control 8.9%, uncultured 7.3%; $P < 0.0001$). We also found significantly greater numbers of SCID repopulating cells (SRC) detected 3 and 9 weeks following transplantation in the Delta1$^{ext\text{-}IgG}$ cultured cells compared to control cultured or non-cultured cells. Relevant to anticipated administration of cultured cord blood units together with a second non-cultured unit in clinical trials, we determined the relative contribution to engraftment of co-infused expanded versus non-manipulated cells in immunodeficient mice, using tissue culture bags as a closed system. We found increased human engraftment in mice that received co-infusions of cultured and uncultured cells compared to either unit alone. Moreover, studies demonstrated that both units contributed to the observed human engraftment suggesting absence of cross-immunologic rejection.

These data demonstrate the ability to ex vivo expand UCB repopulating cells using a clinically relevant Notch ligand-based closed culture system and suggests clinical evaluation of this approach to provide more rapid engraftment to overcome the major disadvantage of UCB transplantation.

25

CD133 HEMATOPOIETIC STEM CELLS IN CORONARY ISCHEMIA

Laughlin, Mary. Case Western Reserve University, Cleveland, OH.

Current stent and bypass therapies in coronary ischemia are limited by problems with restenosis and microvessel disease and innovative strategies are needed. CD133$^+$ hematopoietic stem cells (HSC) are a focus of intensive research in attempt to augment therapeutic neovasculogenesis in response to vascular ischemia. Bone marrow or mobilized peripheral blood from adults has been primarily used as a stem cell source in these laboratory studies and early clinical trials. These studies have involved heterogenous cell populations infused and it is unclear what specific cell populations home to sites of ischemia and mediate neoangiogenesis. This proposal outlines laboratory and clinical phase II studies incorporating a specified CD133$^+$ HSC cell dose administered via intracoronary injection for patients with chronic ischemia. The hypotheses underlying our work are that CD133$^+$ HSC, rather than mature terminally differentiated endothelial cells, are the critical cell population that home to sites of vascular injury in response to inflammatory signals and mediate new vessel formation, primarily via stromal cells and endothelial cells present in the ischemic vascular bed. This presentation will include a brief summary of our ongoing studies focused on determination of vasculogenic function and immunogenicity of marrow-derived enriched AC133$^+$ HSC vs non-selected mononuclear cells, including in vivo functional readouts with Doppler flow measurements and histologic studies after human cell infusions in a NOD.SCID hind-limb ischemia study model. We are testing purified CD133$^+$ HSC cell populations in this model including in vivo tracking to determine whether infused human cell populations integrate and function in new vessel formation, and whether infused cell populations persist over time. This presentation will include preliminary data from our ongoing Phase I clinical trial under FDA IND that demonstrates intracoro-